2 × PCR Solution

PrimeSTAR™ HS (Premix)

Code No. R040A

Shipping at − 20°C

Size: 500 μ I \times 5

Stored at − 20°C

(for 100 PCR reactions)

Storage: Excessively repeated freeze-thaw cycles may decrease the enzyme activity.

Lot No.

Expiry Date:

Description: PrimeSTAR™ HS (Premix) is an optimized mixture composed of PrimeSTAR™ HS DNA Polymerase, which is a high-fidelity DNA polymerase developed by Takara Bio Inc, reaction buffer and dNTP mixture as 2-fold concentration. As this product offers quick preparation of reaction mixture and reduction of contamination risk, it is also useful for high-throughput application.

PrimeSTAR^m HS DNA Polymerase has a matchless proof reading activity due to very strong 3'-5' exonuclease activity, and besides its amplification efficiency is higher than that of Tag DNA Polymerase.

Content:

PrimeSTAR $^{\rm M}$ HS DNA Polymerase * :1.25 units/25 μ I dNTP Mixture :2 \times conc. ; ea. 0.4 mM

PrimeSTAR™ Buffer :2 × conc.; including 2 mM Mg²⁺

* Specification of PrimeSTAR™ HS HS DNA Polymerase (Cat.#R010A)

Unit definition: One unit is the amount of the enzyme that will incorporate 10 nmol of dNTP into acid-insoluble products in 30 minutes at 74°C with activated salmon sperm DNA as the template-primer.

Reaction mixture for unit definition:

100 mM Tris-HCl (pH8.3 at 37°C)

10 mM KCl 2 mM MgCl₂ 6 mM (NH₄)₂SO₄ 0.1% TritonX-100

200 μ M each dATP, dGTP, dCTP

100 μM [³H]TTP 0.001% BSA

0.4 mg/ml activated salmon sperm DNA

Purity: Nicking, endonuclease and exonuclease activity were not detected after incubation of 0.6 $\,\mu$ g of supercoiled pBR322 DNA, 0.6 $\,\mu$ g of $\,\lambda$ -Hind III digest with 10 units of this enzyme for 1 hour at 74°C .

Applications: DNA amplification by Polymerase Chain Reaction (PCR).

PCR product: A significant percentage of PCR product obtained using PrimeSTAR™ HS DNA polymerase will possess blunt-ends. Thus, obtained PCR products can be directly cloned into blunt-end vectors. (If necessary, phosphorylate PCR products before cloning.)

PCR test: Good enzyme performance was confirmed by robust single fragment DNA PCR amplification using both λ DNA (amplified fragments: 8, 10, 12, 15 kb) and Human genomic DNA (amplified fragment: 0.5, 1, 2, 4, 6, 8 kb) as templates.

General reaction mixture for PCR (total 50 μ I):

 $\begin{array}{lll} \text{PrimeSTAR}^{\text{\tiny M}} \text{ HS (Premix)} & 25 \ \mu\text{I} \\ \text{Primer 1} & 0.2\text{-}0.3 \ \mu\text{ M (final conc.)} \\ \text{Primer 2} & 0.2\text{-}0.3 \ \mu\text{ M (final conc.)} \\ \text{Template} & <200 \ \text{ng} \\ \text{Sterilized distilled water} & \text{up to 50 } \mu\text{I} \end{array}$

Recommended template amount

PCR conditions:

98°C 10 sec. 55°C 5 sec.or 15 sec. 30 cycles
72°C 1 min./kb 30 cycles
or
98°C 10 sec. 68°C 1 min./kb

Notice: Because this enzyme possesses an extremely high priming efficiency, use of a short annealing time is strongly recommended. Please refer to the product manual supplied with this product for detailed cycling recommendations.

NOTICE TO PURCHASER: LIMITED LICENSE

[P1] PCR Notice

Use of this product is covered by one or more of the following US patents and corresponding patent claims outside the US: 5,079,352, 5,789,224, 5,618,711, 6,127,155 and claims outside the US corresponding to US Patent No. 4,889,818. The purchase of this product includes a limited, non-transferable immunity from suit under the foregoing patent claims for using only this amount of product for the purchaser's own internal research. No right under any other patent claim (such as the patented 5' Nuclease Process claims in US Patents Nos. 5,210,015 and 5,487,972), no right to perform any patented method, and no right to perform commercial services of any kind, including without limitation reporting the results of purchaser's activities for a fee or other commercial consideration, is conveyed expressly, by implication, or by estoppel. This product is for research use only. Diagnostic uses under Roche patents require a separate license from Roche. Further information on purchasing licenses may be obtained by contacting the Director of Licensing, Applied Biosystems, 850 Lincoln Centre Drive, Foster City, California 94404, USA.

[L15] Hot Start PCR

Licensed under U.S. Patent No. 5,338,671 and 5,587,287 and corresponding patents in other countries.

[M54] PrimeSTAR® HS DNA Polymerase

This product is the subject of the pending U.S. patent application and its foreign counterparts

U.S. Patent 5,436,149 for LA Technology is owned by TAKARA BIO INC.

Note

This product is intended to be used for research purpose only. They are not to be used for drug or diagnostic purposes, nor are they intended for human use. They shall not to be used products as food, cosmetics, or utensils, etc.

Takara products may not be resold or transfered, modified for resale or transfer, or used to manufacture commercial products without written approval from TAKARA BIO INC.

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2 × PCR Solution

PrimeSTAR® HS (Premix)

Code No. R040A

Shipping at − 20°C

Size: 500 μ I \times 5

Stored at − 20°C

(for 100 PCR reactions)

保存:過剰に凍結融解を繰り返すと活性が低下する場合がありますのでご注意ください。

Lot No. (英文面をご覧ください。)

品質保証期限: (英文面をご覧ください。)

●製品説明

PrimeSTAR® HS(Premix)は、タカラバイオが独自に開発した High-Fidelity PCR 酵素、PrimeSTAR® HS DNA Polymerase と反応用バッファー、dNTP Mixture をあらかじめ 2 倍濃度で混合したプレミックス PCR 酵素である。反応液調製の手間が大幅に減少し、また、コンタミネーションの危険性も軽減するため、ハイスループットな実験系にも有用である。本製品に使用している PrimeSTAR® HS DNA Polymerase は非常に強力な 3′ → 5′ exonuclease 活性を有し、DNA 合成において抜群の校正力を示す一方、Tag DNA Polymerase に優る高い増幅効率も示す。

●内容

PrimeSTAR® HS DNA Polymerase * :1.25 units/25 μI dNTP Mixture :2 × conc. ; 各 0.4 mM

PrimeSTAR® Buffer :2 × conc.; 2 mM Mg^{2 +}を含む

* PrimeSTAR® HS DNA Polymerase (製品コード R010A)

○活性の定義

活性化サケ精子 DNA を鋳型/プライマーとして用い、下記の活性測 定用反応液中にて 74℃において、30 分間に 10 nmol の全ヌクレオチドを酸不溶性沈殿物に取り込む活性を 1U とする。

○活性測定用反応液組成

100 mM Tris-HCl 緩衝液 (pH8.3 at 37℃)

10 mM KCI 2 mM MgCl₂ 6 mM (NH4) 2SO4 0.1% TritonX-100 各 200 μM dATP, dGTP, dCTP

100 μM [³H] TTP 0.001% BSA

0.4 mg/ml 活性化サケ精子 DNA

○純度

- 1. 10 U の本酵素と 0.6 µg の λ-Hind III 分解物とを 74℃、1 時間反応させても DNA の電気泳動パターンに変化は起こらない。
- 10 U の本酵素と 0.6 µg の supercoiled pBR322 DNA とを 74℃、1 時間反応させても DNA の電気泳動パターンに変化 は起こらない。
- ●用途 Polymerase Chain Reaction (PCR) 法による DNA 増幅

● PCR 産物

 $PrimeSTAR^{\circ}$ HS DNA Polymerase を用いて増幅した PCR 産物のほとんど は平滑末端である。したがって、その PCR 産物をそのまま(必要に応じてリン酸化を行って)平滑末端のベクターにクローニングすることが可能である。

● PCR 検定

- λ DNA を鋳型とした PCR 反応(増幅産物 8,10,12,15 kb)において 良好な増幅が見られることを確認している。
- 2. ヒト Genomic DNA を鋳型とした PCR 反応 (増幅産物 0.5, 1, 2, 4, 6, 8 kb) において良好な増幅が見られることを確認している。

● PCR 反応例(total 50 µI PCR)

PrimeSTAR® HS(Premix) 25 μ I Primer 1 0.2 \sim 0.3 μ M(final conc.) Primer 2 0.2 \sim 0.3 μ M(final conc.) Template < 200 ng 滅菌蒸留水 up to 50 μ I

※ Template 量の目安

E h Genomic DNA $5\,\mathrm{ng} \sim 200\,\mathrm{ng}$ (< $200\,\mathrm{ng}$)E. coli Genomic DNA $100\,\mathrm{pg} \sim 100\,\mathrm{ng}$ cDNA \ni $\mathcal{I} \supset \mathcal{I} \supset \mathcal{I}$ $1 \sim 200\,\mathrm{ng}$ λ DNA $10\,\mathrm{pg} \sim 10\,\mathrm{ng}$ $\mathcal{I} \supset \mathcal{I} \supset \mathcal{I} \supset \mathcal{I}$ $10\,\mathrm{pg} \sim 10\,\mathrm{ng}$ $\mathcal{I} \supset \mathcal{I} \supset \mathcal{I} \supset \mathcal{I}$ $10\,\mathrm{pg} \sim 10\,\mathrm{ng}$

● PCR 条件

98°C 10 sec. 55°C 5 sec. または 15 sec. 72°C 1 min. /kb 30 cycles or 98°C 10 sec. 68°C 1 min. /kb 30 cycles

> ※本酵素は高いプライミング効率を有しているため、アニーリング 時間を短かく設定することをお勧めします。ここに挙げた条件は あくまで一例ですので、PCR条件の詳細な設定方法については、 添付の取扱説明書をご確認ください。

●注意

本製品は研究用として販売しております。ヒト、動物への医療、臨 床診断用には使用しないようご注意ください。また、食品、化粧品、 家庭用品等として使用しないでください。

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